

Appl. No. *To Be Assigned*; Filed: *Herewith*
 Dkt. No. 1716.0510009; Group Art Unit: *To Be Assigned*
 Inventors: Miettinen-Oinonen *et al.*; Tel.: (202) 371-2600
 Title: Novel Cellulases, the Genes Encoding Them and Uses
 Thereof

ALKO4179: pH-DEPENDENCY OF THE ENDOGLUCANASE ACTIVITY

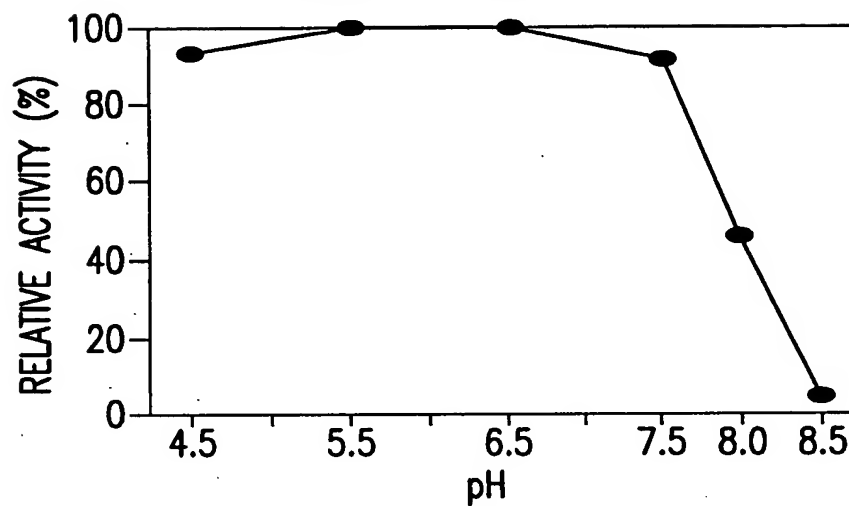


FIG.1A

ALKO4179: TEMPERATURE DEPENDENCY OF THE ENDOGLUCANASE ACTIVITY

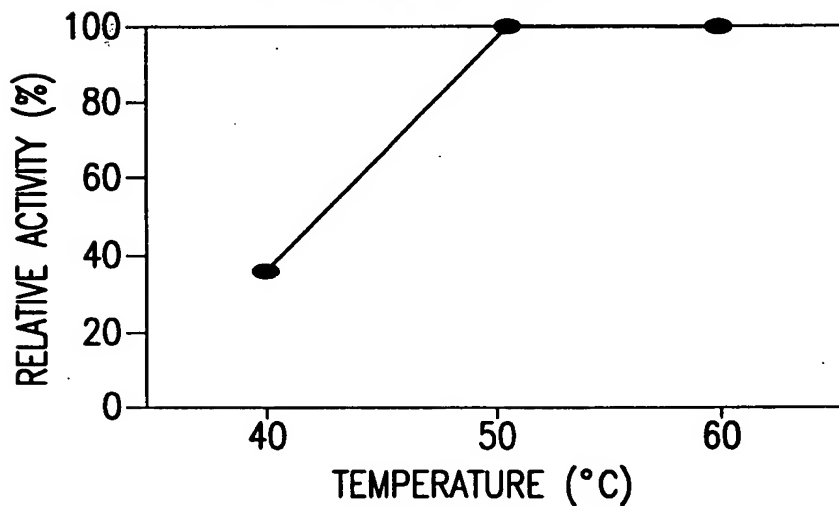


FIG.1B

ALKO4124: pH-DEPENDENCY OF
THE ENDOGLUCANASE ACTIVITY

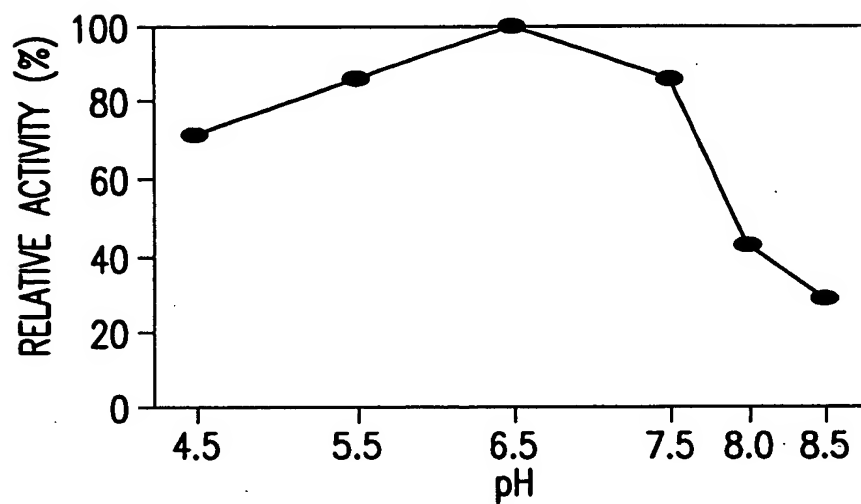


FIG.2A

ALKO4124: TEMPERATURE DEPENDENCY OF
THE ENDOGLUCANASE ACTIVITY

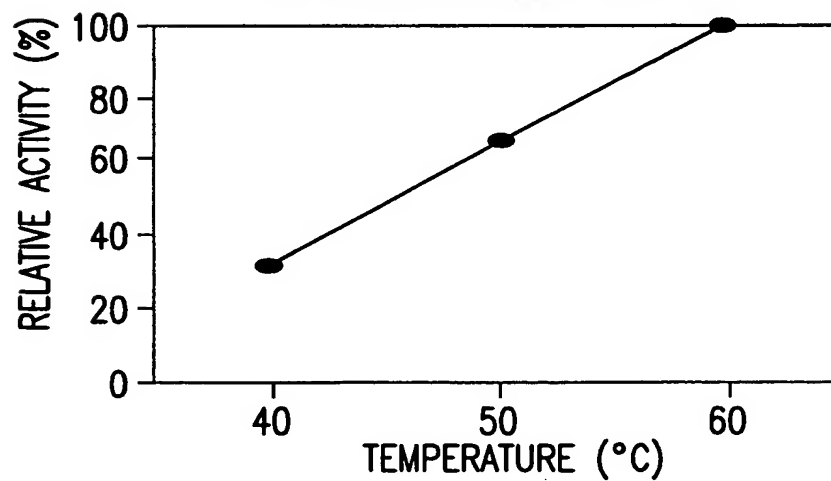


FIG.2B

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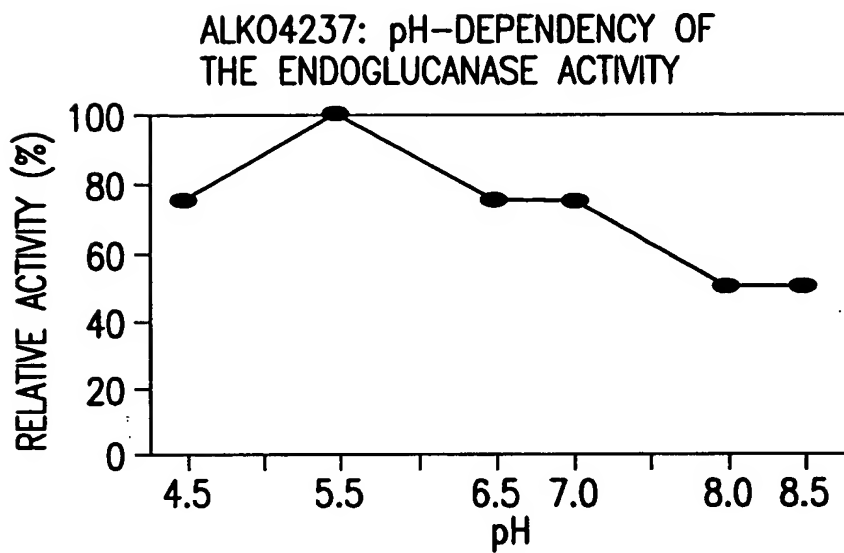


FIG.3A

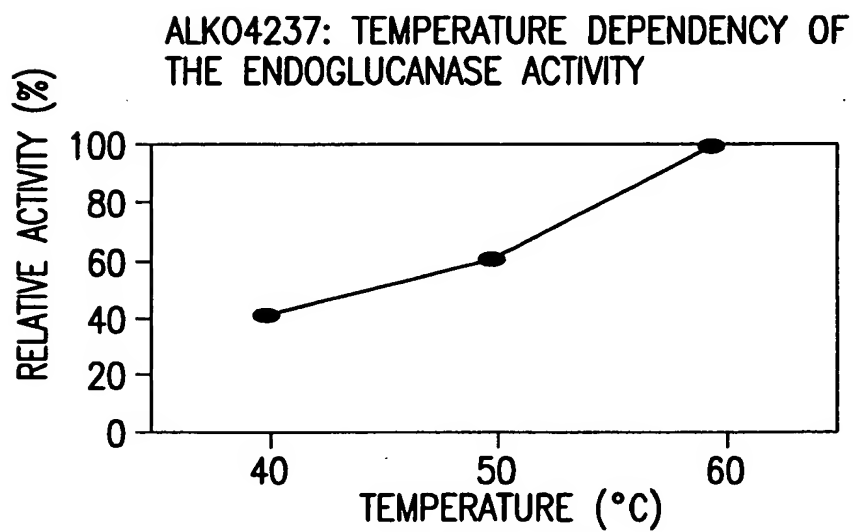


FIG.3B

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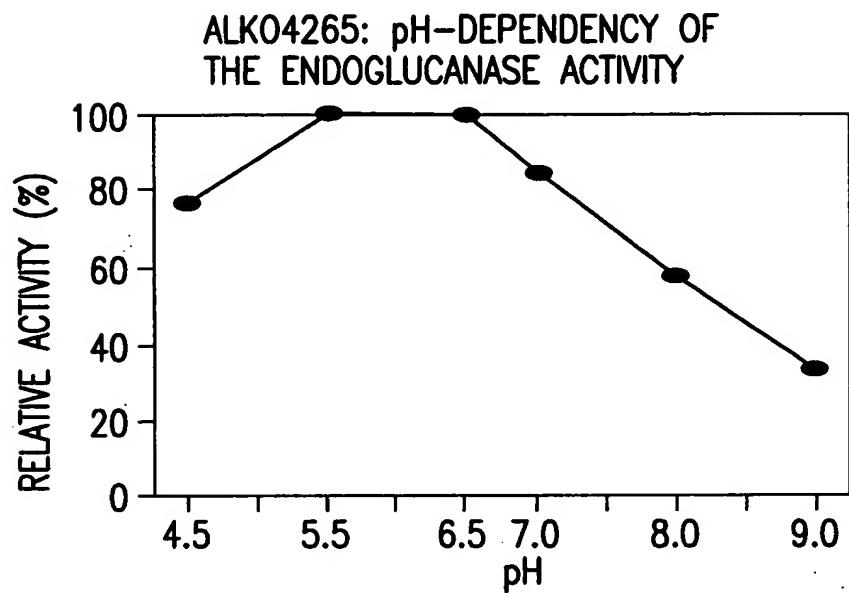


FIG.4A

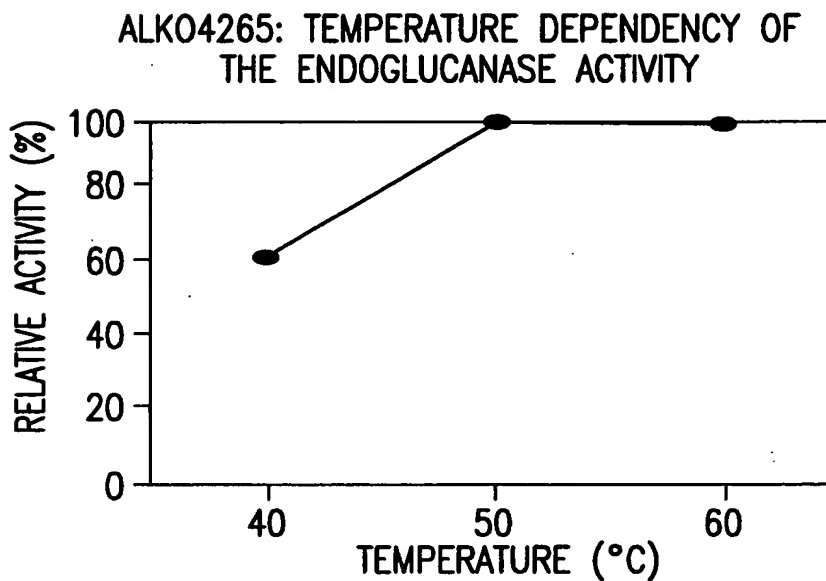


FIG.4B

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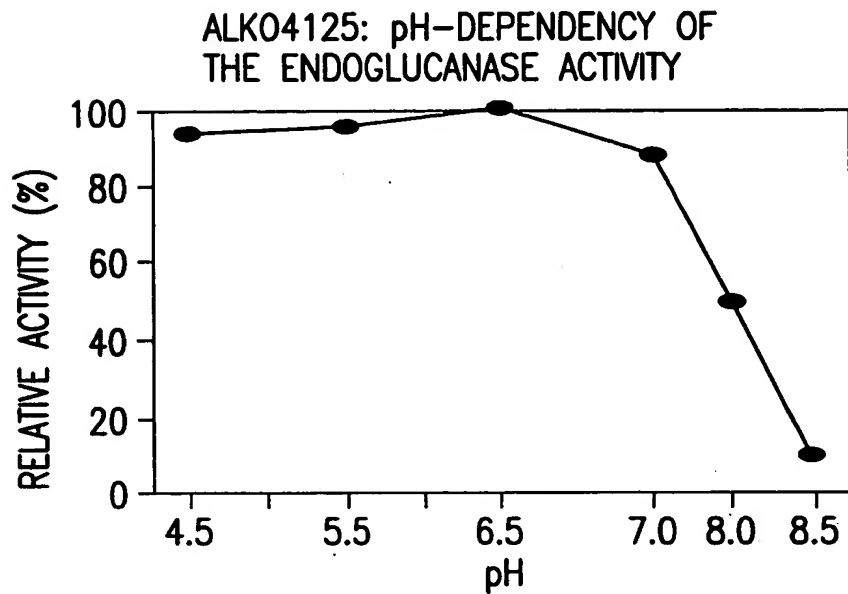


FIG.5A

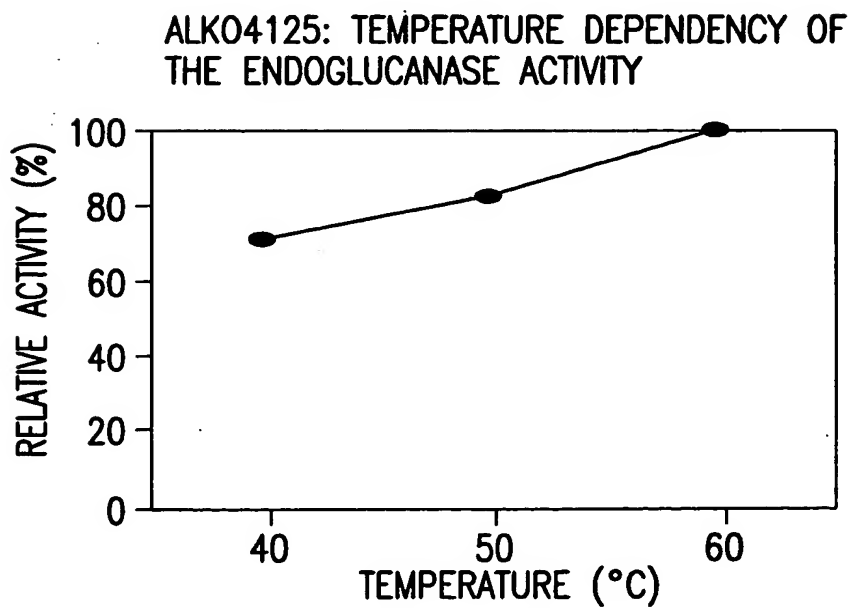


FIG.5B

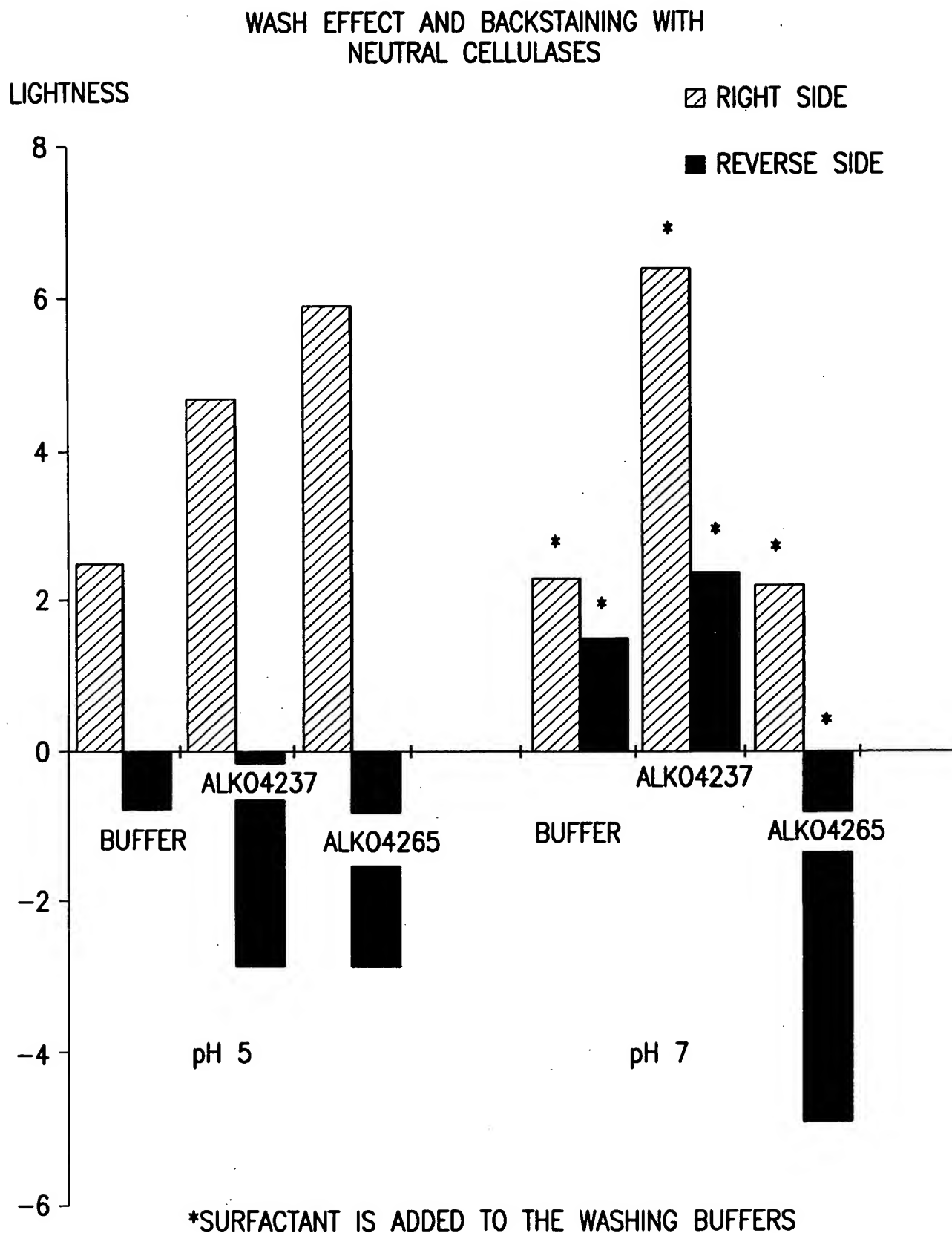
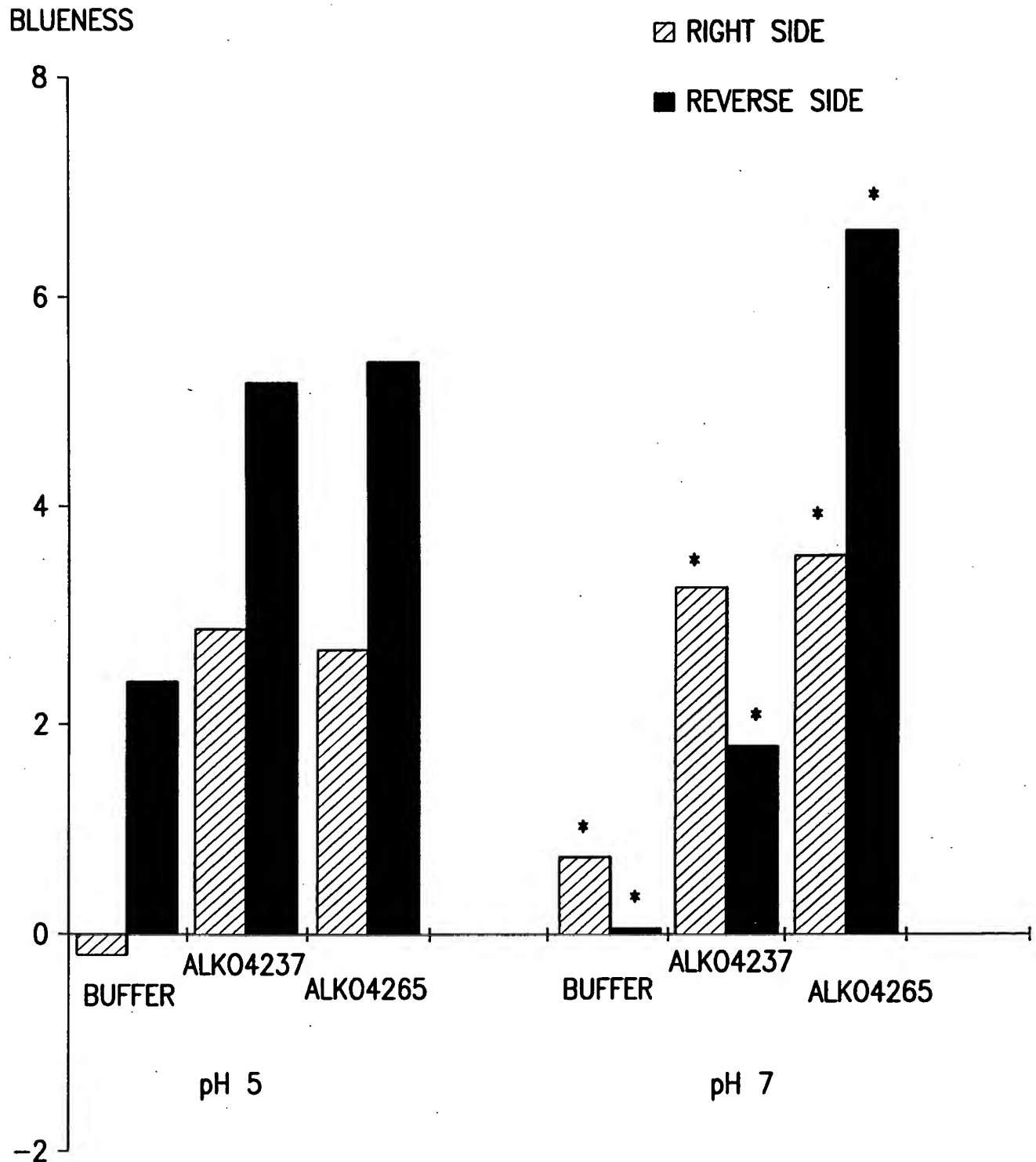


FIG.6A

BLUENESS WITH THE NEUTRAL CELLULASES



*SURFACTANT ADDED TO THE WASHING BUFFERS

FIG.6B

WASH EFFECT AND BACKSTAINING WITH ECOSTONE L

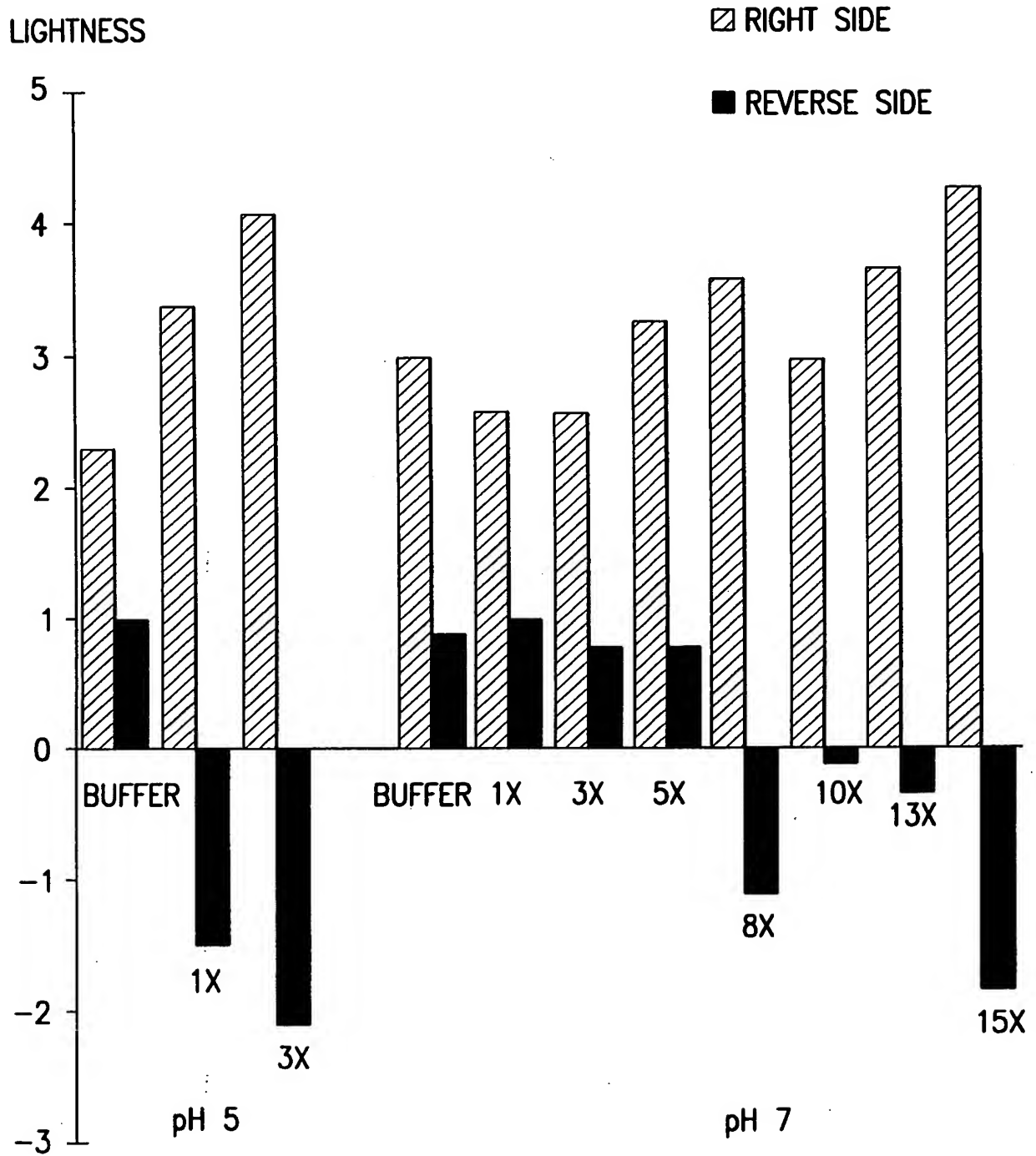


FIG.7A

BLUENESS WITH ECOSTONE L

BLUENESS

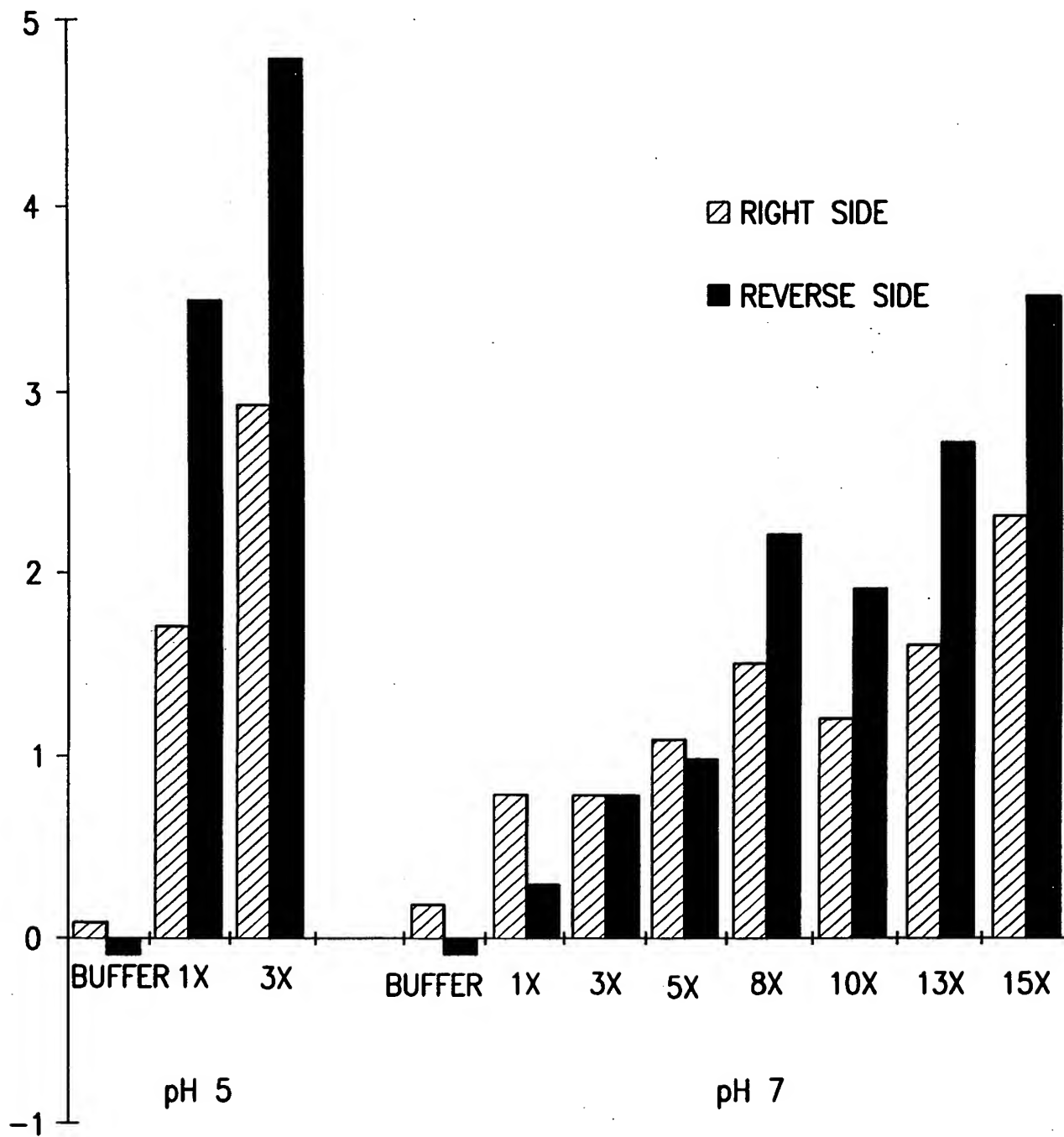


FIG.7B

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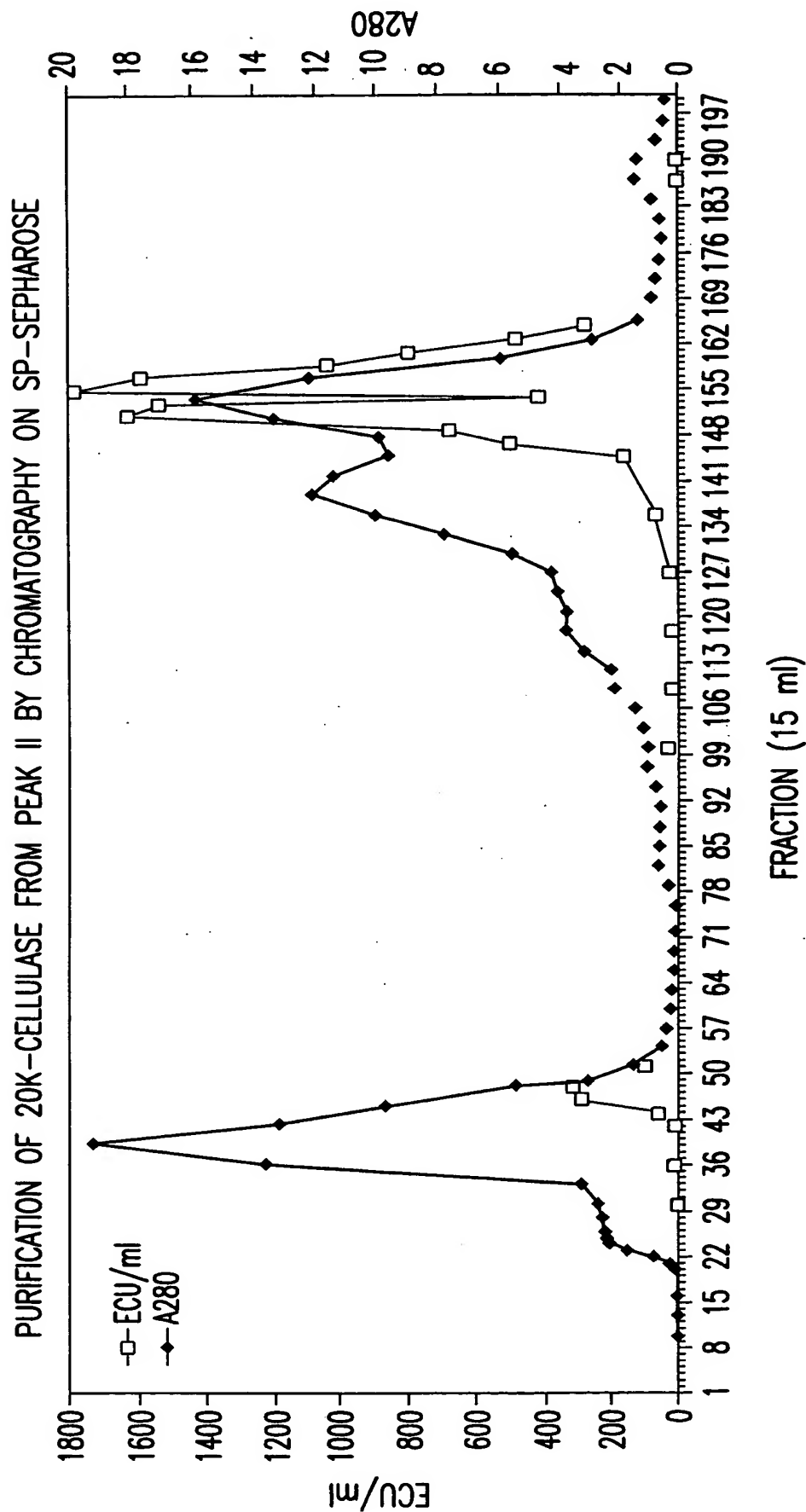


FIG.8

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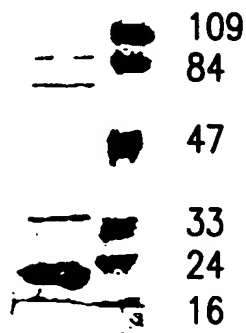


FIG.9A

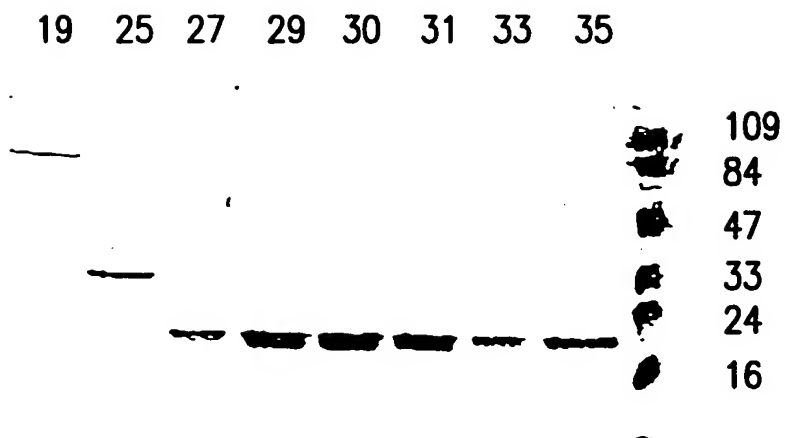


FIG.9B

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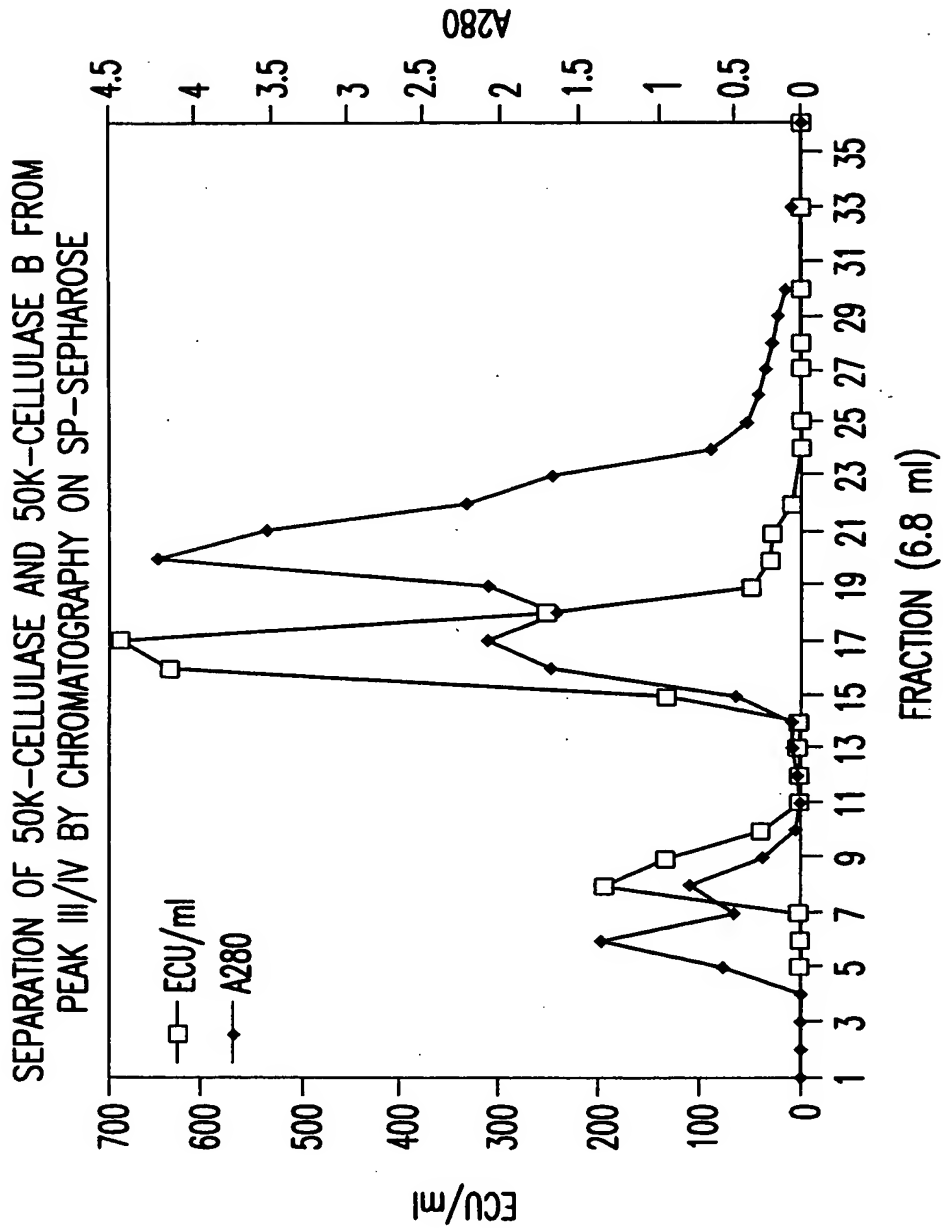


FIG.10

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BEST AVAILABLE COPY

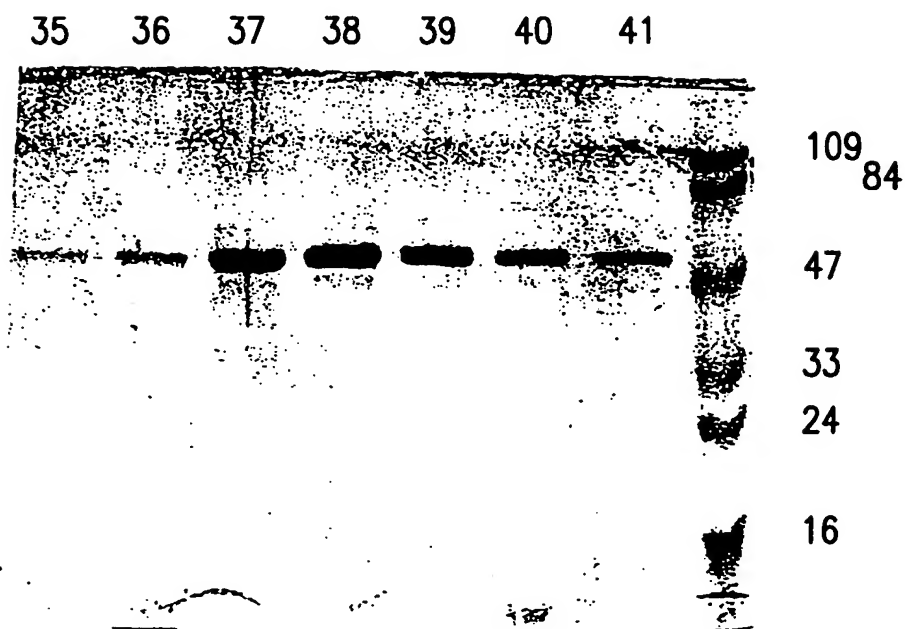


FIG. 11A

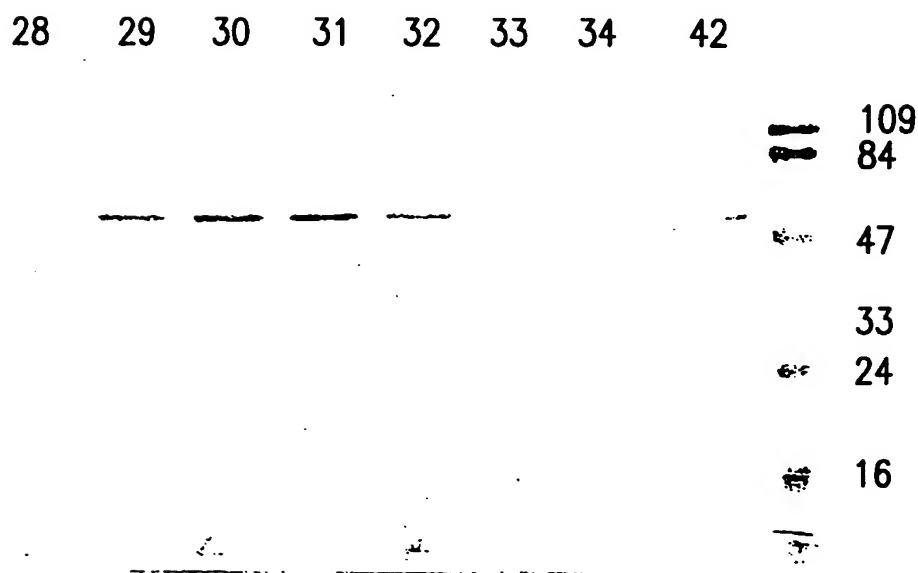


FIG. 11B

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TEMPERATURE DEPENDENCE OF THE
ENDOGLUCANASE ACTIVITY OF 50K-CELLULASE
AT pH 7.0

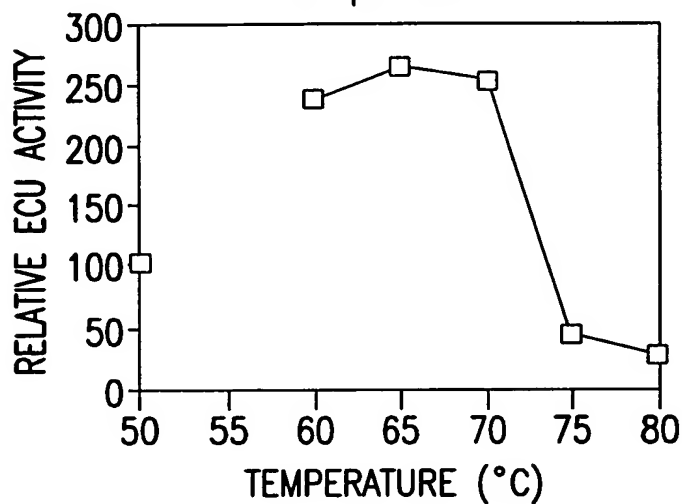


FIG.12

THE pH DEPENDENCE OF THE ENDOGLUCANASE
ACTIVITY OF 50K-CELLULASE AT 50°C AND 70°C

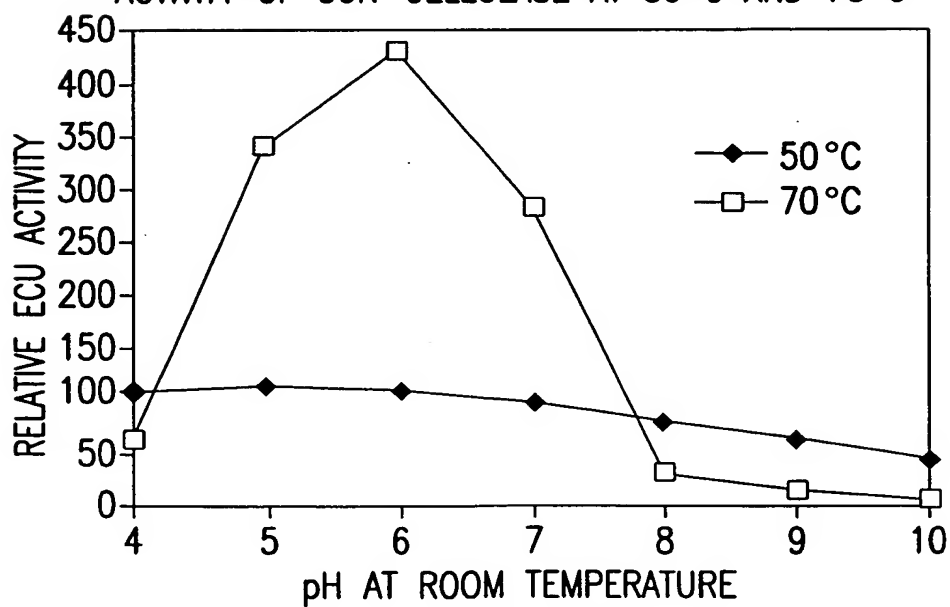


FIG.13

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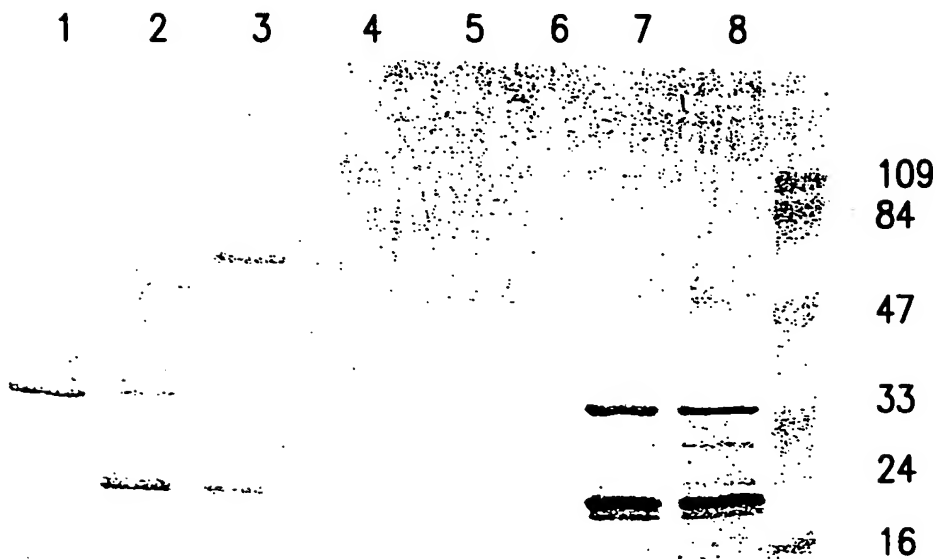


FIG.14

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20K-CELLULASE: TEMPERATURE DEPENDENCE OF
ENDOGLUCANASE ACTIVITY AT pH 7 (10 MIN REACTION
TIMES)

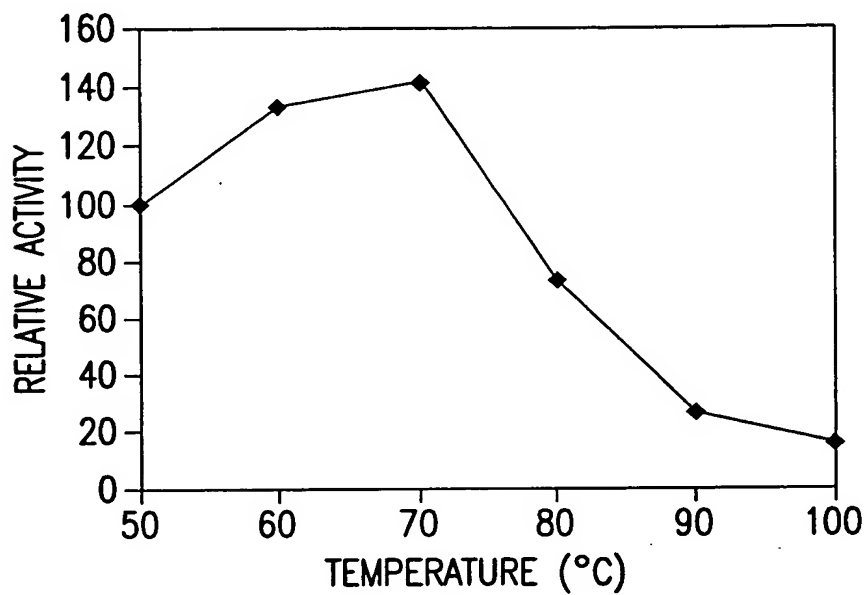


FIG.15

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20K-CELLULASE: pH-DEPENDENCE OF ENDOGLUCANASE
ACTIVITY (10 MIN REACTION TIMES)

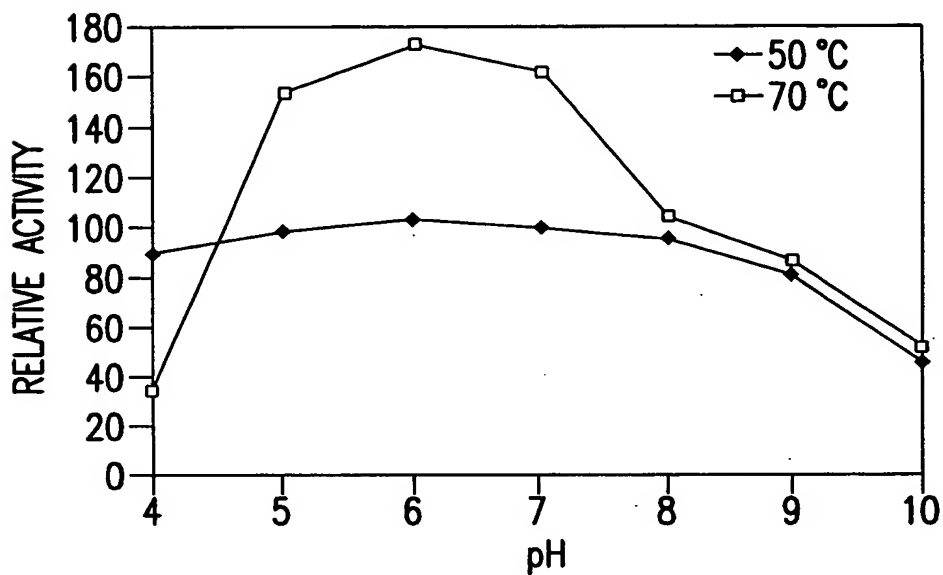


FIG.16A

20K-CELLULASE: pH-DEPENDENCE OF ENDOGLUCANASE
ACTIVITY (60 MIN REACTION TIMES)

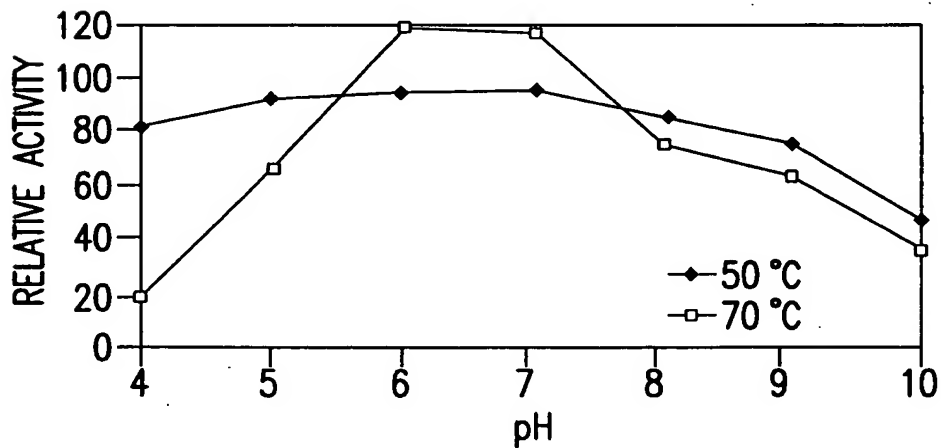


FIG.16B

429
ANGQSTRYWDCCKPSCGWRGKGPVNQPVYS

430
YGGISSR

431
CGWR

432
PSCGWR

433
YWDCCK

439
QECDSFPEPLKPGCQWR

fr 9
RHDDGGFA

fr 14
YWDCCKP

fr 16
GKGPVNQPVYSCDANFQR

fr 17
VQCPEELVAR

fr 28
DWFQNADNPSFTFER

fr 30
TMVVQSTSTGGDLGSNHFDLNIPGGGVGLF

FIG.17

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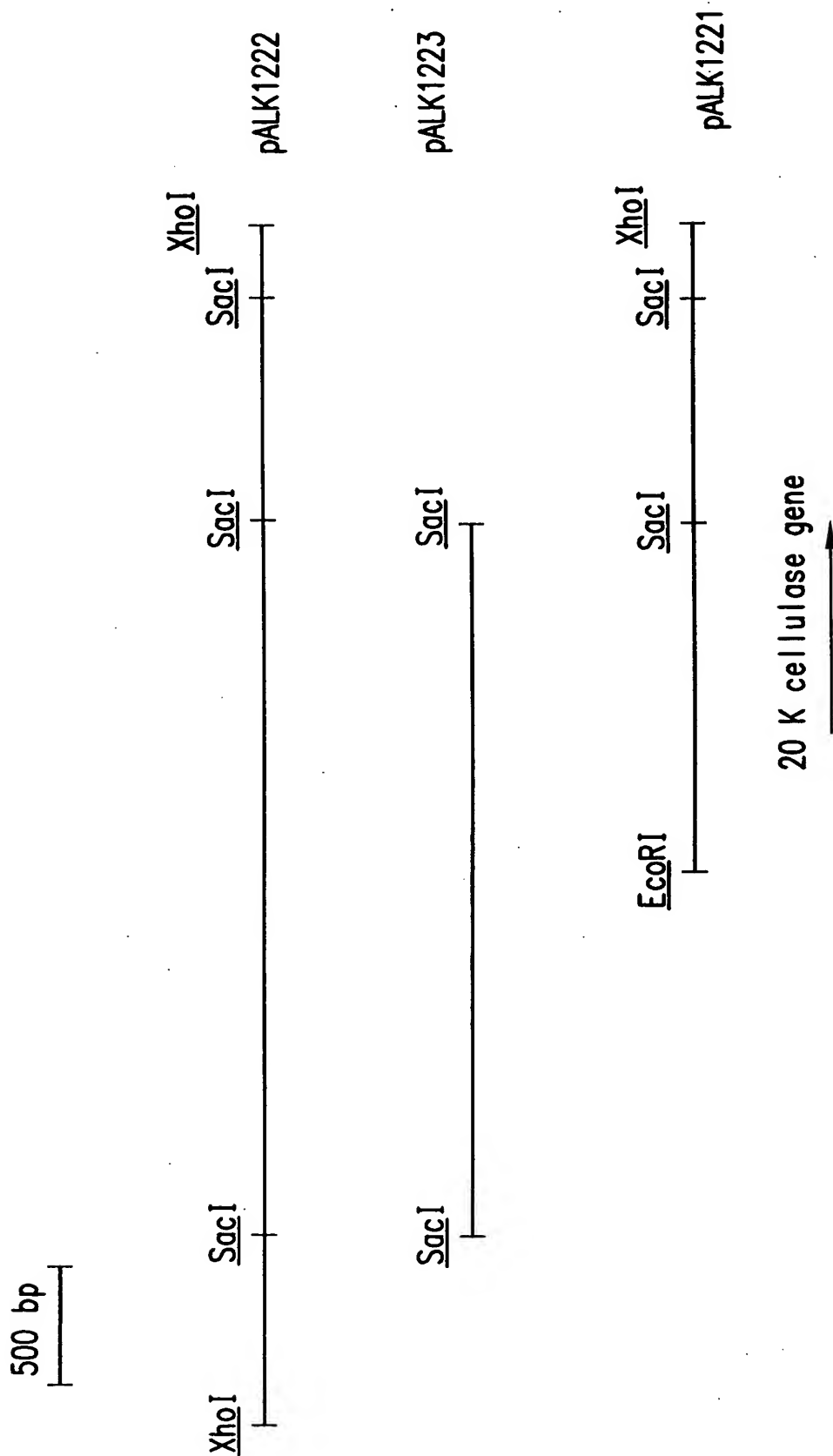


FIG.18

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-30      -10      10      30      50
TCGCCCCCTAACCGAGAACCAAGACTCCAAGAAATGCGGCTCTACTCCCGTTCTCCGGCGCCCTCCTGGCCGCAGCATTGCCCCCTCGGGGCCCTCGCCGCCCAA
      M R S T P V L R A L L A A L P L G A L A A N
70      90      110      130      150
CGGTCAGTCCACGAGgtaactgatcaccgcctcattacgcgtgccgacggaccgcgttcagggtcactgctcaccgcattccagATACTGGGACTGCT
G Q S T R      Y W D C C
170      190      210      230      250
GCAAGCGTCTGTCGGCTGGCGCGGAAAGGGCCCGTGAACCAAGCCCGTCTACTCGTGGCAGCGCAACTTCCAGCGCATCCACGACTTCGATGCCGTCTC
K P S C G W R G K G P V N Q P V Y S C D A N F Q R I H D F D A V S
270      290      310      330      350
GGGCTGCGAGGGCGGCCCGCTTCTCGTGGCGCGACACAGCCCTGGGCCATTAAATGACAACCTCTCTGTACGGCTTCGCGGGGACTGCACCTCAGCGGC
G C E G G P A F S C A D H S P W A I N D N L S Y G F A A T A L S G
370      390      410      430      450
CAGACCGAGGAGTCGTGGTGTGCTGCTACGCGtgtagtgctgtgggcccacgtcggtgattccggagttcagaccactgaccacgagcccgctc
Q T E E S W C C A C Y A

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FIG. 19A

```

470          490          510          530          550
gccagTCTGACCTTTACATCGGGTCCC GTGGCCGGCAAGACCATGGTCCAGTCGACCAAGCGGGCGGCACCTCGGCAGCAACCACTTCGACCTCA
    L T F T S G P V A G K T M V V Q S T S T G G D L G S N H F D L N

570          590          610          630          650
ACATCCCCGGGGGGGTGGCCCTCTTCGACGGGTGCACTCCCCAGTTCGGCGGCCTCCCGGGCGCACGGTACGGCGGCATCTCGTCGGCGCCAGGAGTG
    I P G G G V G L F D G C T P Q F G G L P G A R Y G G I S S R Q E C

670          690          710          730          750
CGACTCGTTCGCCGAGCGGCTCAAGCCCGGTGCCAGTGGCGCTTCGACTGGTCCAGAACGCCGACACCCGTCCTTTACCTTCGAGCGGGTCCAGTGC
    D S F P E P L K P G C Q W R F D W F Q N A D N P S F T F E R V Q C

770          790          810          830          850
CCCGAGGAGCTGGTCGCTCGGACCGGCTGCAGGGCGCCACGACGCGGGCTTCGCCGCTCTTCAAGGCCCCCGAGCGCCTGATCCGTTTTTGGGCAGTGTC
    P E E L V A R T G C R R H D D G G F A V F K A P S A *

870          890
CGTGTGACGGCAGCTACGTGGAACGACCTGGAGCTC

```

Fig. 19B

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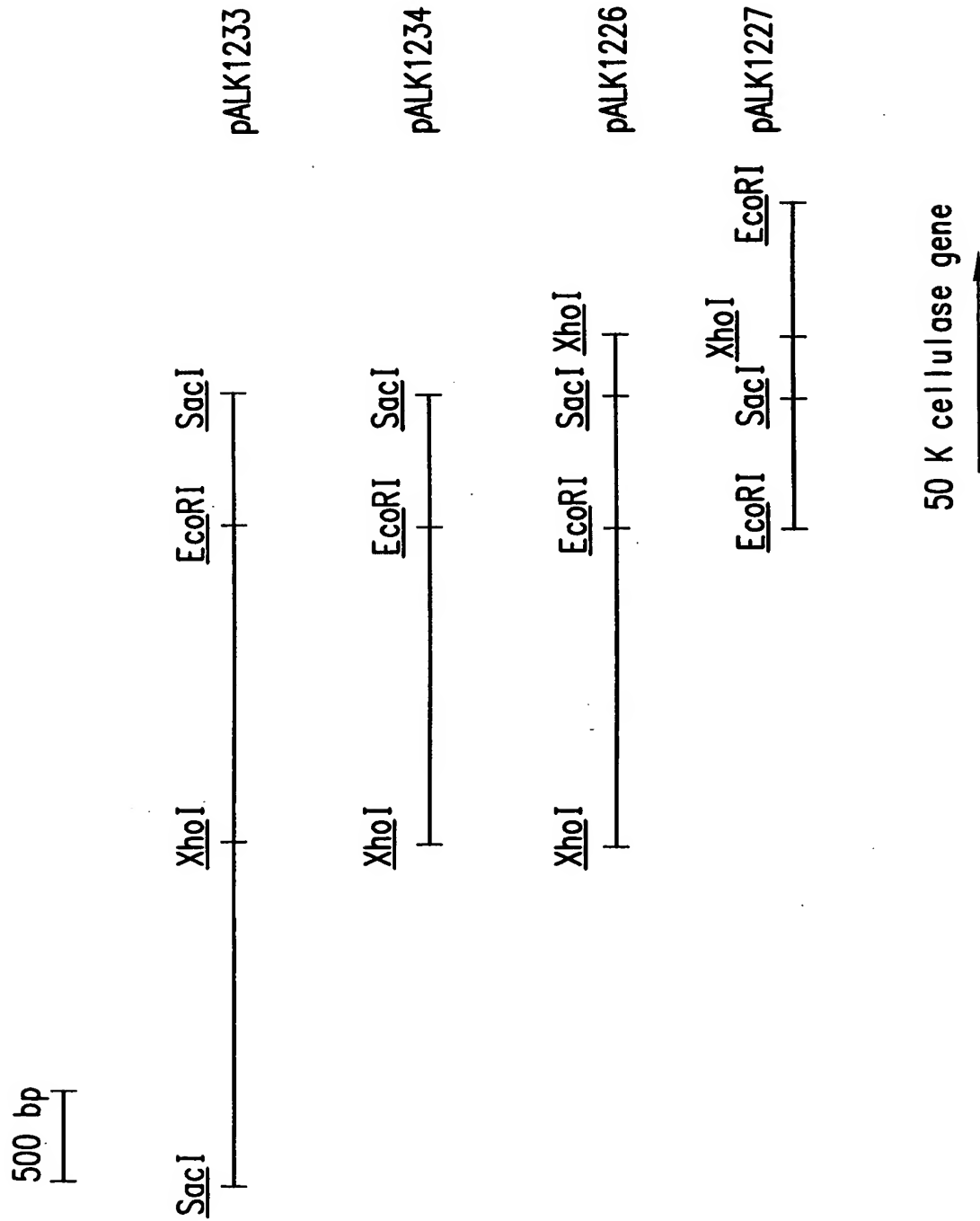


FIG.20

-230	-210	-190	-170	-150
GAATTCGGGGGTTGCCAGGAGTCGTACAGGGGTGGGTGGAGGGGGATGGAGGGGATGGAGAAGAAAGCATATATGGGACGTTTGTGCTC				
-130	-110	-90	-70	-50
GCCGGCTCCCTCTGCCACGTTCCCTTGCCCTGCGTGGGTGTTGTTGGTCTTCCCTTCACCATCCGACAAACCAACCTGCTGCGGGTGAACCTCGCA				
-30	-10	10	30	50
GAGCGCCTTCGGACGACGACAGACAGCGCACCATGACTCGCAACATCGCCCTGCTCGGGCGCGGTGCGGGCTCCTGGGCCTCGCCACGGCCAGAAGC				
70	90	110	130	150
M T R N I A L L G A A S A L L G L A H G Q K P				
CGGGCGAGACGCCCGAGGTGCACCGCAGCTGACGACGTTCCGGTGACCAAGCGGACGGGTGCCAGCCGGACCAACTACATTGTGCTGGACTCGCT				
170	190	210	230	250
G E T P E V H P Q L T T F R C T K A D G C Q P R T N Y I V L D S L				
GTGCAACCGGTGGACAACGACTACAACTGCGGGGACTGGGGGCAGAAAGCCCAACGCGACGGGTGCCCGGACGTCGAGTCGTGCGGCGGC				
270	290	310	330	350
S H P V H Q V D N D Y N C G D W G Q K P N A T A C P D V E S C A R				
AACTGCATCATGGAGGGGTTGCCCGGACTACAGCCAGCAGCGGTACGACGAGCGACAGCTGCTGGCCTGCAGCAGCTCGTCGACGGCCGCTCGTCA				
N C I M E G V P D Y S Q H G V T T S D T S L R L Q Q L V D G R L V T				

FIG.21A

```

370          390          410          430          450
CGCGGGCGTCTACCTGCTCGACGAGACCGGACCGCTACGAGATGATGCACCTGACCGGCCGAGGATTCACCTTTGAGGTCGACGCCACCAAGCTGCC
P R V Y L L D E T E H R Y E M M H L T G Q E F T F E V D A T K L P

470          490          510          530          550
CTGCGGCATGAACAGCGCCCTCTACCTGTCCGAGATGGACCGACCGGCGCCCGGAGCGAGCTCAACCCCGCGGTGCCTACTACGGCACCGGCTACTGCG
C G M N S A L Y L S E M D P T G A R S E L N P G G A Y Y G T G Y C

570          590          610          630          650
GACGCCCAGTGTCTGTGACGCCATTTCATCAACGGCATTgtgagtggtcccttggccccccccctgaaaaatagatgtacctgggtgctaaccgccggg
D A Q C F V T P F I N G I

670          690          710          730          750
tgtcgaccacaaacagGGCAACATCGAGGGCAAGGGCTCGTGTGCAACGAGATGGACATCTGGGAGGCCAACTCGCGGGCGACGCACGTGGCGCGGCAC
G N I E G K G S C C N E M D I W E A N S R A T H V A P H

770          790          810          830          850
ACGTGCAACCAAGCGGTCTGTACATGTGCGAGGGCGCGGAGTGCAGTACGACGGCGTGTGCGACAAGGACGGGTGCGGGTGGAAACCGTACCGGGTCA
T C N Q T G L Y M C E G A E C E Y D G V C D K D G C G W N P Y R V N

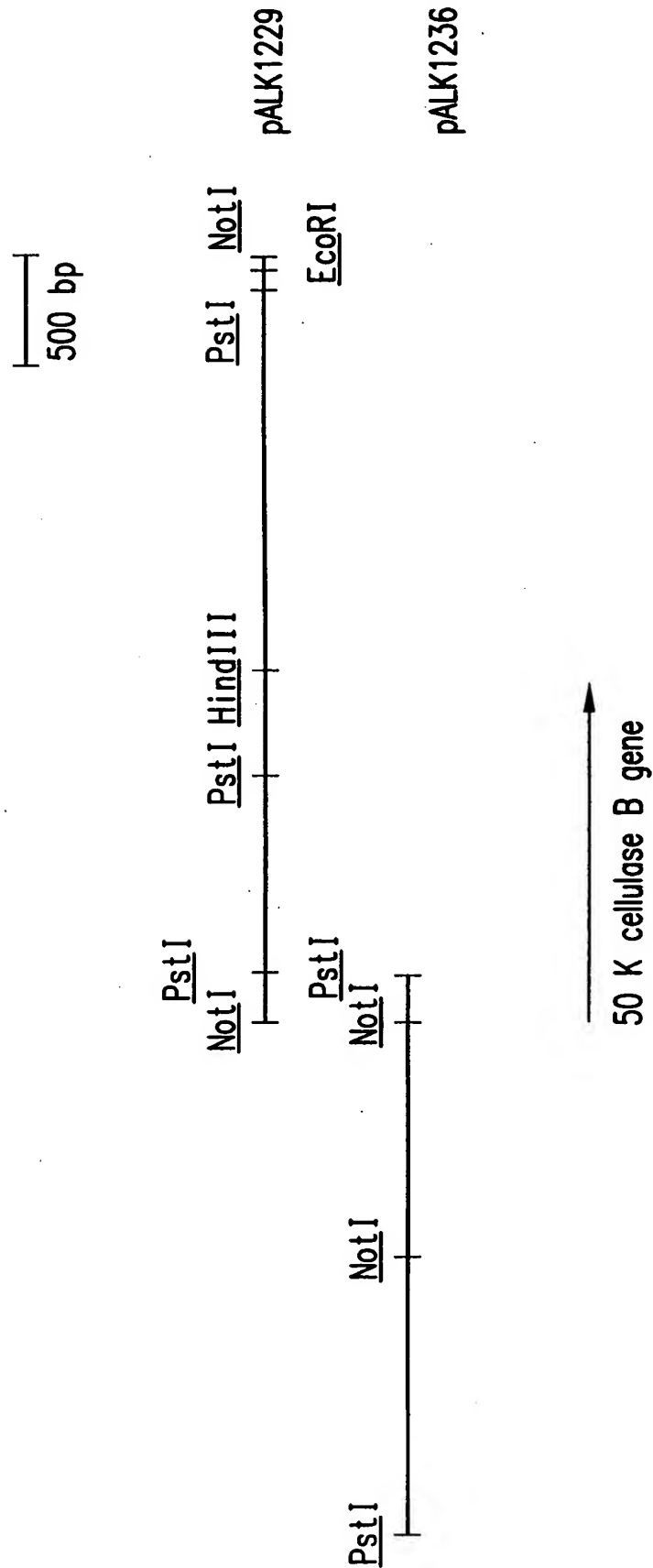
870          890          910          930          950
ACATCACCAGCTACTACGGCAACTCGGACCGGTTCCGGCTCGACACGGGGGGCCCTTCACCGTGGTGACGCAGTTCCTCGGGCCGACGCCGAGGGCCGGCT
I T D Y Y G N S D A F R V D T R R P F T V V T Q F P A D A E G R L

```

FIG. 21B

970	990	1010	1030	1050
CGAGAGCATCCACCGGCTGTACGTGCAGGACGGCAAGGTGATCGAGTCGTACGTGACGCGCGGGCCTGCCCCGGACCGACTCGCTCAACGACGAG				
E S I H R L Y V Q D G K V I E S Y V V D A P G L P R T D S L N D E				
1070	1090	1110	1130	1150
TTCTGCGCCGCCACGGGGCGCGGCTACCTCGACCTCGGGCGGACCGGGGCATGGGCGACGCCATGACGGCGGCAITGGTCTGGCCATGAGCATCT				
F C A A T G A A R Y L D L G G T A G M G D A M T R G M V L A M S I W				
1170	1190	1210	1230	1250
GGTGGACGAGTCCGGCTTCATGAACCTGGCTCGACAGCGGGCGAGGGCCCTGCCTGCCCGACGAGGGCGACCCCAAGAACATTGTCAAGGTCGAGCC				
W D E S G F M N W L D S G E A G P C L P D E G D P K N I V K V E P				
1270	1290	1310	1330	1350
CAGCCCCGAGGTACCTACAGCAACCTGCGCTGGGGCGAGATCGGGTCGACCTTTGAGGCCGAGTCCGACGACGACGGCGGCGGCGGACGACTGCTAGATA				
S P E V T Y S N L R W G E I G S T F E A E S D D G D G D D C *				
1370	1390	1410	1430	1450
ACTAACTAGTGGCGGAAAGGGCGGGGATGCGTAACCTACATACAGCCCGGAGTTGTTTGAGTGTAGAGTATTGAGCTTTCGATGTTAGTTGAGTG				
1470	1490	1510	1530	1550
GAATGGAAAATTCCGGTCTTTGCCCGGTTGGTGATAAACAATAGTCGGCTGGTGCAATTGTGACACTTCAATTGCGCTGTTGGCTTGGTGACAGACA				
1570	1590	1610	1630	1650
CGGCAGCGTCGATGACCCGACACCCAGAATAATTGCGATGGTTGATTANTGTTATTGTGCTTTAAATCGGAGGCTGATGCTCATCTCTCGAATTC				

FIG.21C



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-150      -130      -110      -90      -70
CCCGGTCTGGAGACGGGGAGCGCGCCAGCGACGAGGATAAGAGGGACGACCGCGCCTCCGAGCCAGGCCCCAGGACAGCAGGAGAACTCGCCACGCGC

-50      -30      -10      10      30
AAGCAGCACGCCCGGATCGACAGTGTCCCGCTCTGCCCCACAGCACTCTGCAACCATGATGATGAAGCAGTACCTCCAGTACCTCGCGGCGCGGCTGCCGCT
      M M M K Q Y L Q Y L A A A L P L

50      70      90      110      130
CGTCGGCCTCGCCGCGCGCCAGCGCGCTGGTAACGAGACGCCCGGAGAACCCCGCTCACCTGGCAGAGGTGCACGGCCCCGGGCAACTGCCAGACC
V G L A A G Q R A G N E T P E N H P P L T W Q R C T A P G N C Q T

150      170      190      210      230
GTGAACGCCGAGGTGTCATTGACGCCCAACTGGCGGTGGTGACGACGACGACAACATGCAGAACTGCTACGACGGCAACCACTGGACCAACGCCTGCAGCA
V N A E V V I D A N W R W L H D D N M Q N C Y D G N Q W T N A C S T

250      270      290      310      330
CCGCCACCGACTGCGCTGAGAAGTGCATGATCGAGGGTGCCGGCGACTACCTGGGCACCTACGGCGCCTCGACCGGGCGACGCCCTGACGCTCAAGTT
A T D C A E K C M I E G A G D Y L G T Y G A S T S G D A L T L K F

350      370      390      410      430
CGTCACCAAGCAGGATACGGCACCAACGTGGCTCGCGCTTCTACCTCATGAACGGCCCGGACAAAGTACCAGATGTTCAACCTCATGGGCAACGAGCTT
V T K H E Y G T N V G S R F Y L M N G P D K Y Q M F N L M G N E L

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FIG.23A

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450          470          490          510          530
GCCTTTGACGTGACCTCTCGACCGTCGAGTGGCGCATCAACAGCGCCCTGTACTTCGTGCGCCATGGAGGACGGCGGCATGGCCAGCTACCCGAGCA
A F D V D L S T V E C G I N S A L Y F V A M E E D G G M A S Y P S N

550          570          590          610          630
ACCAGCGCGCGCCGGTACGGGCACGTGGGGTgagttgagctcgcgtttgtttcgagtcgcaacgaggcactttctggcgccggctaactctctcgattc
Q A G A R Y G T G

650          670          690          710          730
ctccgacagTACTGCGATGCCCCAATGCGCTCGTGATCTCAAGTTCGTGGCGGCAAGGCCAACATTGAGGGCTGGAAGTCGTCCACCAGCGACCCCAACG
Y C D A Q C A R D L K F V G G K A N I E G W K S S T S D P N A

750          770          790          810          830
CTGGCGTCGGCCCGTACGGCAGCTGTGCGGTGAGATCGACGTCGTgagtcgagaccgtccaccaggtttcggaatgcggggtggaatttcgcggct
G V G P Y G S C C A E I D V W

850          870          890          910          930
aacggagcacccccagGGAGTCGAATGCCTATGCCCTTCGCTTCACGCCGCGACGGTGCACGACCAACGAGTACCACGCTCGCGAGACCACCAACTGCG
E S N A Y A F A F T P H A C T T N E Y H V C E T T N C G

950          970          990          1010          1030
GTGGCACCTACTCGGAGGACCGCTTCGCCGGCAAGTGGACGCCAACGGTGGACTACAAACCCCTACCGCATGGGCAACCCCGACTTCTACGGCAAGGG
G T Y S E D R F A G K C D A N G C D Y N P Y R M G N P D F Y G K G

1050          1070          1090          1110          1130
CAAGACGCTCGACACCGCGCAAGTTCACgtgctgaccccttgtagcgcaacctttctctgcctgcctggacacactgaaactgacacgtcgttttcg
K T L D T S R K F T

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FIG.23B

1150	1170	1190	1210	1230
gctgcagCGTCTCGTCTCCGGCTTCGAGGAGAACAGGCTCTCCAGTACTTCATCCAGGACGGCCGAAGATCGAGATCCGCGCCGCGGAGGGCAT				
	V V S R F E E N K L S Q Y F I Q D G R K I E I P P T W E G M			
1250	1270	1290	1310	1330
GCCAACAGCAGGAGATCACCCCGAGCTCTGCTCCACCATTGTCGATGTGTTCAACGACCGCAACCGCTTCGAGGAGGTTCGGCGGCTTCGAGCAGCTG				
	P N S S E I T P E L C S T M F D V F N D R N R F E E V G G F E Q L			
1350	1370	1390	1410	1430
AACAAAGCCCTCCGGGTTCCTCGTCTCGTCAATGTCATCTGGGACGACgtaagtaccgcgcgacctccctagccacacaagccgcgcgagggc				
	N N A L R V P M V L V M S I W D D			
1450	1470	1490	1510	1530
agcccatcgctgctgtaacacgagaccgttcgtagCACTACGCCAACATGCTCTGGCTCGACTCCATCTACCCGCCGAGAGGAGGCCAGCCCGGCG				
	H Y A N M L W L D S I Y P P E K E G Q P G A			
1550	1570	1590	1610	1630
CCGCCCGTGGCGACTGCCCCACGGACTCGGGTGTCCCCCGCGAGGTTCGAGGCTCAGTTCCTCCCGACGCGtaagacttgcccccgacccccaaagcttccactt				
	A R G D C P T D S G V P A E V E A Q F P D A			
1650	1670	1690	1710	1730
ctggatgccgaatgctaaacgcgaaacagCCAGGTGCTCTGGTCCAAACATCCGCTTCGGCCCATCGGCTCGACCTACGACTTCTAAGCCGGTCCATGC				
	Q V V W S N I R F G P I G S T Y D F *			
1750	1770	1790	1810	1830
ACTCGAGCCCTGGGCGCGTCAAGCCCGCCACCTCCCTCGCGGAAACTCTCCGTGCGTCCGGGGCTCCAAAGCATTTTGGCCTCAAGTTTTTTCGTTTC				

FIG.23C

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 Inventors: Miettinen-Oinonen *et al.*; Tel.: (202) 371-2600
 Title: Novel Cellulases, the Genes Encoding Them and Uses
 Thereof

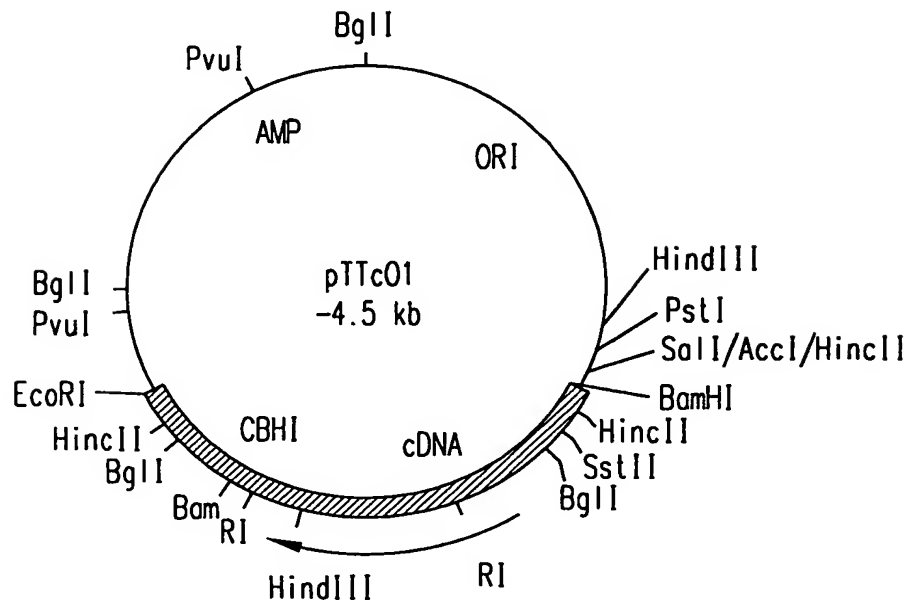


FIG.24

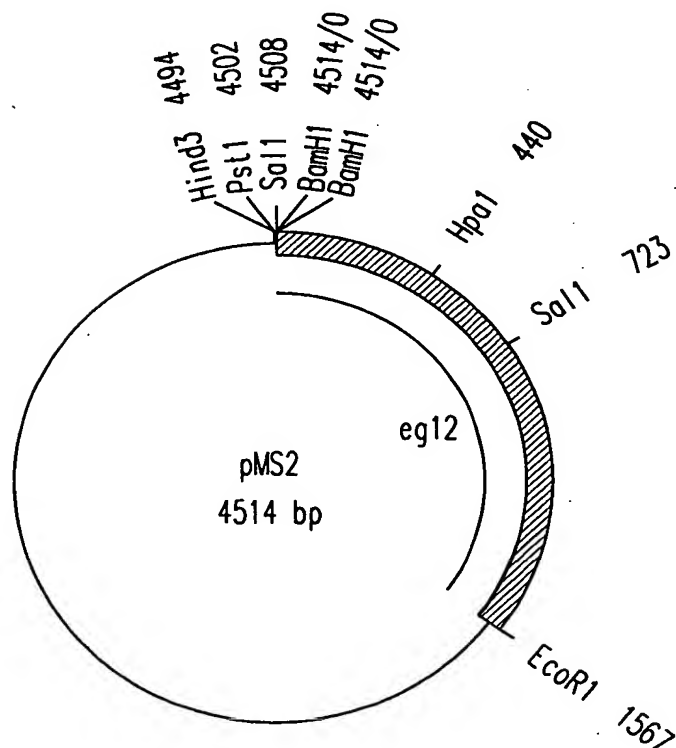


FIG.25

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 Dkt. No. 1716.0510009; Group Art Unit: *To Be Assigned*
 Inventors: Miettinen-Oinonen *et al.*; Tel.: (202) 371-2600
 Title: Novel Cellulases, the Genes Encoding Them and Uses
 Thereof

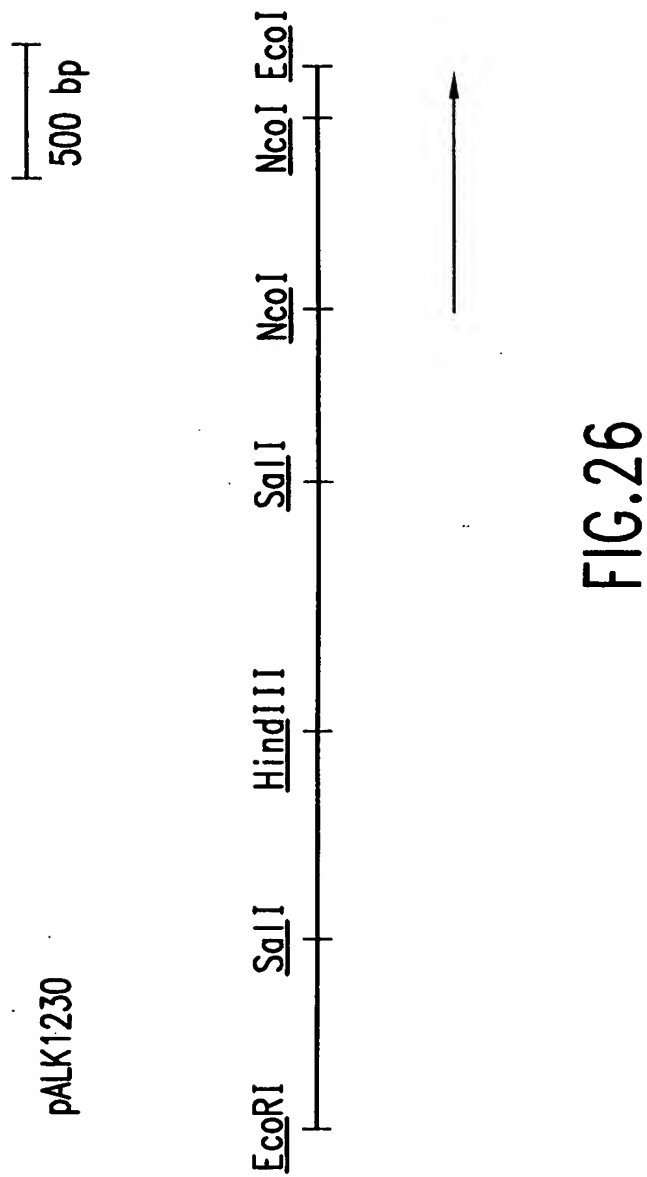
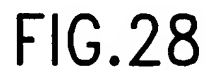


FIG.26

10	30	50	70	90
CCATGGACGGAAGTGGACGTCTTCTGCCCCGAGCTGAAGACCCAGAGCATCCAGACCGGCAACCAAGTGCACCCAGGAGATGAAGGTCTACGAGAACAAT				
110	130	150	170	190
TGACGGCTGGCTCGACAGCCTGCCCGGGAACGTCCCCCATCACCGGTCCGACGCCGGCTCTGGTAAGTCAAAGAGATGATGCCTACCTACCTTCCCACCT				
210	230	250	270	290
TCCCACCCAGCCGCAAAATACCTTTCTCCCTCCCCGTGCCCGTATCTTTCAACGCCCGGAGACTGACAGACCCGCTCGTCCCAGGCGGCAACCCCGGCA				
310	330	350	370	390
ACGGCGGGGCGCAGCAACCCGGGCAACGGGCGGGCGGGGCTGCACCGTCCAGAAAGTGGGGCCAGTCCGGCGGCATCGGCTACTCGGGCTGCACCACTG				
410	430	450	470	490
CAAGGCCGGCTCGACCTGCCCGGCCAGAAACGAGTACTACTCGCAGTGCCTGTAAAGCGGCCGTGGCTAGGTGGCCGAGCGGGGGGTTTCTTCATTGG				
K A G S T C P A Q N E Y Y S Q C L *				
510	530	550	570	590
TTGAGCAAATAGAACAGGAATTCGGGCTCGTTGGCAGCGGCGCGCGGGGATGGTGTGTACAATTCAGACCTCAGTACCGAGGGAACCTGGAAAGGA				
610	630	650	670	690
GTCAGTCTGCTTGACGGAGGCTGGCTGCCCGGTGGCGGGGCTGGCAAGGTAGATAGCCCTTCATTGCTGTAAGTGTCTATATACCTCTGCACATT				
710	730	750	770	790
TGCAGCCCCATGGTGTGAACAACAAGTGAACAGGCTTCCAGTTCAGCCTCGCGCAATTGTCAGGATATCCTTGGTCCAICTATATGTAATGGGCATGAGC				
810	830	850	870	
GAGTCGAGAAATGTACCGGAAAAATCGTAGTGACCTGGCGACTGGCGGCTTCTACCAACCGTAGGATTGAAGTGAATCTCGAATTC				

FIG.27



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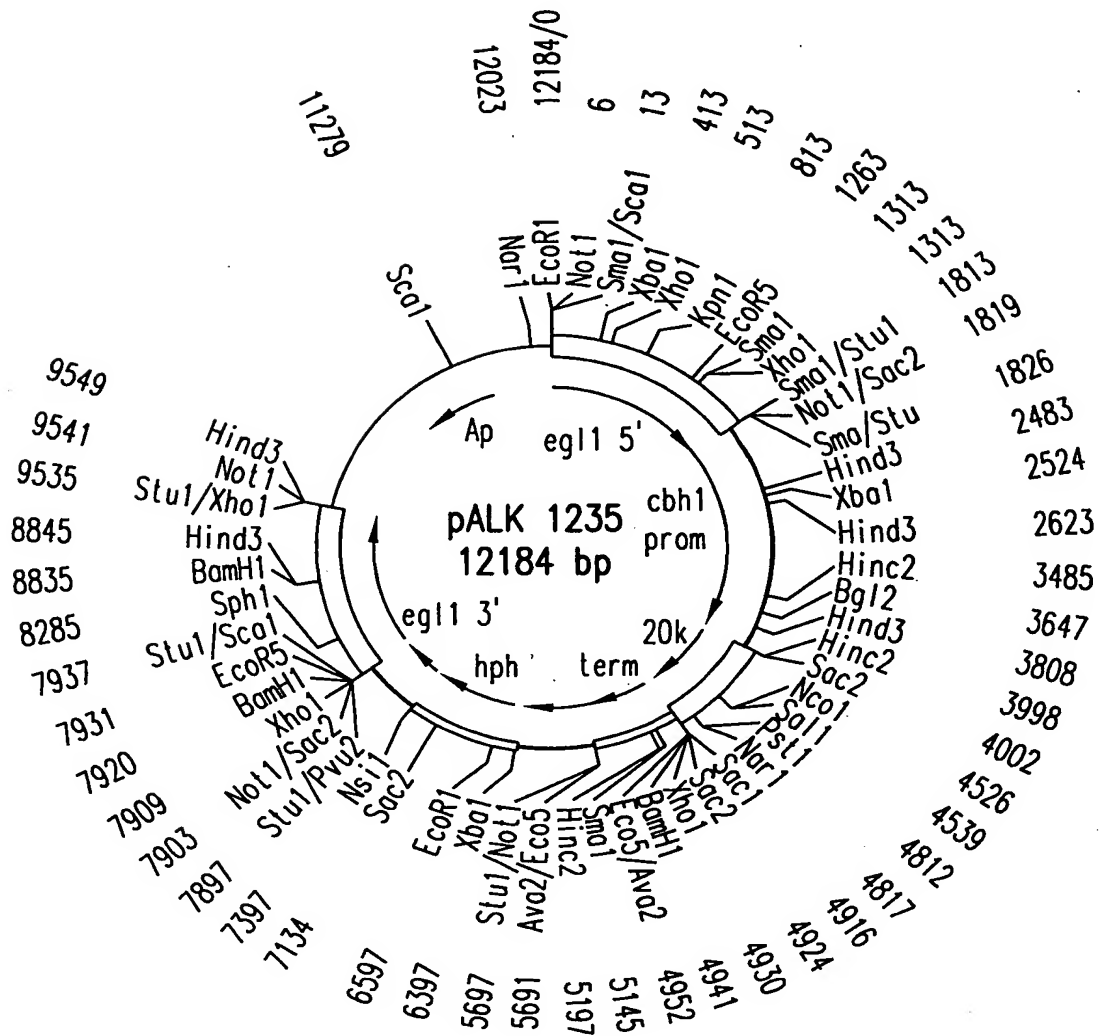


FIG.29

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Thereof

1 2 3 4 5 6

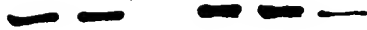


FIG.30

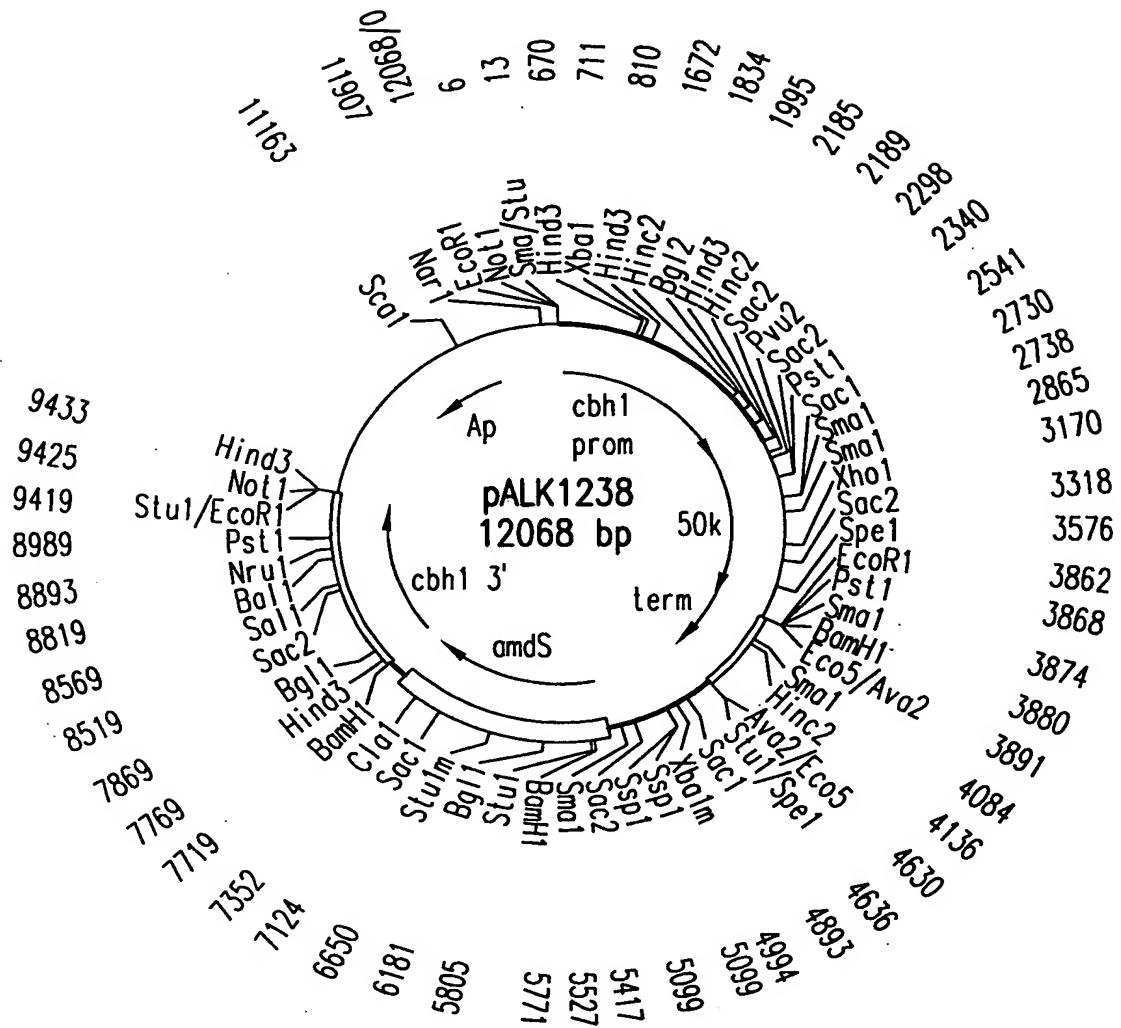


FIG.31

